PATENT

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE
BEFORE THE BOARD OF PATENT APPEALS AND INTERFERENCES

In re Application of:	)	Confirmation No. 8424
	)	
Cooper, Mark J., et al.	)	Group Art Unit 1633
	)	
Serial No. 10/656,192	)	Examiner: Long, Scott
	)	
	)	Atty. Dkt. No. 003659.00029
Filing Date: September 08, 2003	)	

FOR: LYOPHILIZABLE AND ENHANCED COMPACTED NUCLEIC ACIDS

BRIEF ON APPEAL

U.S. Patent and Trademark Office
Randolph Building
401 Dulany Street
Alexandria, VA 22314

Sir:

Appellants filed the Notice of Appeal on March 27, 2009. Charge the fee for filing this Brief and any other required fees to our Deposit Account No. 19-0733.

REAL PARTY IN INTEREST

The real party in interest in this application is Copernicus Therapeutics, Inc.

RELATED APPEALS AND INTERFERENCES

There are no related appeals or interferences.

STATUS OF CLAIMS

Claims 1-5, 8-14, 17-19, 26, 28, 30-31, 34-35, 38-40, 51-55, 58-70, 73-82, 103-104, 106-107, 114-115, and 122 are pending. Claims 6-7, 15-16, 20-25, 27, 29, 32-33, 36-37, 41-50, 56-57, 71-72, 83-102, 105, 108-113, 116-121, and 123 are cancelled.[1]

STATUS OF AMENDMENTS AFTER FINAL REJECTION

No amendments were filed after the final rejection.

SUMMARY OF CLAIMED SUBJECT MATTER

Claim 1

A composition is provided which contains complexes of a nucleic acid molecule and one or more polycation molecules. (Page 3, line 13-14). The complexes have particular properties and they are made by a particular method. The properties of the complexes include:

- unaggregated (page 2, line 15-16);
- rod-shaped (when visualized by transmission electron microscopy) (page 7, line 27-29);
- 10-20 nm diameter (when visualized by transmission electron microscopy) (page 16, line 1-2);

[1] All pending claims are rejected. A restriction and election of species has focused the examination on the species: acetate counterion, cDNA nucleic acid molecule, and CK15-60P10 polycation. CK15-60P10 is a polyamino acid polymer of one N-terminal cysteine and 15-60 lysine residues, with a molecule of polyethylene glycol having an average molecular weight of

- nucleic acid is condensed (page 22, line 26-28);
- colloidally stable in normal saline (page 8, line 11-16).

The method of making the complexes comprises: mixing the nucleic acid molecule and the polycation molecules. (Page 2, line 5-6). Prior to the mixing step, the polycation molecules have a counterion which is acetate, bicarbonate, or chloride (page 3, line 1-2).[2]

Claim 8

The subject matter is similar to claim 1 but differs in two respects:

- the nucleic acid molecule encodes at least one functional protein (page 3, line 29);
- the polycation has a nucleic acid binding moiety through which it complexes to nucleic acid (page 28, line 6-7).

Claim 17

The subject matter is similar to claim 1 but differs in two respects:

- the nucleic acid molecule is a double-stranded cDNA (page 4, lines 8-9);
- the nucleic acid molecule encodes at least one functional protein. (Page 3, line 29)

Claim 26

The subject matter is similar to claim 1 but differs in three respects:

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10 kdal attached to the cysteine residue. Page 2, lines 24-27.

[2] Only the elected species of acetate is under examination.

- the complexes are described as soluble rather than unaggregated (page 5, line 20-21);
- acetate is the only recited counterion to the polycation (page 9, line 20-24);
- the step of mixing is performed at a salt concentration sufficient for compaction of the complexes. (Page 3, lines 14-19.)

Claim 28

The subject matter is similar to claim 1 but differs in three respects:

- the complexes are described as soluble and without aggregates (page 5, line 20-21);
- acetate is the only recited counterion to the polycation (page 9, line 20-24);
- the step of mixing is performed in the absence of added salt (page 5, line 20-21).

GROUNDS OF REJECTION TO BE REVIEWED

I. Whether Hanson[3] anticipates claims 1, 2, 8, 9, 11, 12, 17, 18, 26, 28, 30, 34, 38, 53, 65, 78, and 103 under 35 U.S.C. §102(b)

II. Whether Hanson, Park[4], and Schacht[5] render claims 3, 10, 19, 31, 35, 51-53, 63-65, 67, 68, 76-78, and 104 obvious under 35 U.S.C. §103(a)

[3] U.S. 5,844,107

III. Whether Hanson, Park, and Mao[6] render claims 58-62, 66, 73-75, 79-82, and 122 obvious under 35 U.S.C. §103(a)

IV. Whether Hanson, Park, Schacht, and Kwoh[7] render claims 4-5, 13-14, 39-40, 54-55, 69-70, 106-107, 114-115 obvious under 35 U.S.C. §103(a)

ARGUMENT

I. Hanson U.S. 5,844,107 does not anticipate claims 1, 2, 8, 9, 11, 12, 17, 18, 26, 28, 30, 34, 38, 53, 65, 78, and 103 under 35 U.S.C. §102(b)

A. Legal standard for anticipation

Section 102(b) states:

> A person shall be entitled to a patent unless -
> ....
>
> (b) the invention was patented or described in a printed publication in this or a foreign country or in public use or on sale in this country, more than one year prior to the date of application for patent in the United States....

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[4] U.S. 6,177,274
[5] WO1998/19710
[6] *J. Controlled Release 70:* 399-421 (2001)
[7] *Biochimica et Biophysica Acta 1444:* 171-190 (1999)

To anticipate a claim, the reference must teach every element of the claim. "A claim is anticipated only if each and every element as set forth in the claim is found, either expressly or inherently described, in a single prior art reference." *Verdegaal Bros. v. Union Oil Co. of California,* 814 F.2d 628, 631, 2 U.S.P.Q.2d 1051, 1053 (Fed. Cir. 1987).

B. Claimed subject matter

The rejected claim group is represented by claim 1, discussed above. Claim 1 contains both product limitations and product-by process limitations. The product limitations related to complexes contained within the claimed compositions include:

- unaggregated;
- rod-shaped;
- 10-20 nm diameter;
- nucleic acid is condensed;
- colloidally stable in normal saline.

The process limitations are:

- mixing the nucleic acid molecule and the polycation molecules.
- the polycation molecules have a counterion which is acetate, bicarbonate, or chloride prior to the mixing step.

C. The Rejection

The U.S. Patent and Trademark Office has characterized the rejection as based on **inherency**. "The examiner has asserted throughout the prosecution history that rods and toroids are both **inherently** present in the nucleic acid formulations like those of Hanson and the instant application."[8] Similarly, "[s]ince Hanson et al., (U.S. 5,844,107) practices the process described in claim 1, therefore the product (i.e., rod-shaped, 10-20 nm diameter, condensed, single nucleic acid molecule complexes) is **inherently** produced.... Because claim 1 is a product-by-process claim and Hanson et al., teaches this process, the examiner asserts that such a rod-shaped complex is formed."[9]

In addition, the U.S. Patent and Trademark Office asserts that Hanson describes products that **inherently** fulfill the product limitations recited in the claims. Although the U.S. Patent and Trademark Office concedes that Hanson does not explicitly describe the complexes recited, it asserts that the complexes which Hanson does describe, relaxed toroids, are interchangeable forms which spontaneously interconvert to condensed rods.[10]

D. Reference teachings

1. Hanson, U.S. 5,844,107

a. complexes

The final office action points to Hanson's teaching of nucleic acid complexes at col. 62, lines 51-57, which reads:

> Electron microscopic results have been indicated as follows: the association of the polycation with the DNA results in aggregation into complexes of increased size (>60 nm) (Aggregated); the structure resulting from the condensation are rod-like relaxed toroids of

[8] Final office action mailed October 27, 2008 at page 3, lines 17-20, emphasis added
[9] Advisory Action mailed March 09, 2009 at page 2, lines 10-11 and 15-16, emphasis added
[10] Final office action at page 4, lines 1-14

> increased size (Relaxed); polycation binding results in proper condensation (toroids <30 nm in diameter) (Condensed). The number of properly condensed structures (toroids) per microscopic field has not been determined. There is approximately 3-fold variation in the number of toroids visible in the EL with different preparations of DNA complex.

Thus Hanson teaches three components in a mixture:

1. aggregate complexes of increased size (>60 nm) (Aggregated)
2. rod-like relaxed toroids of increased size (Relaxed)
3. proper condensation (toroids <30 nm in diameter) (Condensed).

b. acetate

The final office action asserts that Hanson teaches the process of the product-by-process limitation, which requires the use of a polycation with acetate as its counterion. Advisory action at page 2, line 6. Hanson (on the U.S. Patent and Trademark Office database) has been electronically searched for every occurrence of "acetate." Each is provided below, with the word acetate highlighted.

- The method of claim 1 wherein diameter of the complex is measured using uranyl **acetate** staining and electron microscopy. Claim 11.
- FIGS. 1B-1G are electronic micrographs (EM). 1B-1D, 1F and 1G are taken at 300,000.times.. The bar in 1D represents 33.3 nm. FIG. 1E was taken at 600,000.times., and the bar is 16.6 nm long. Uranyl **acetate** staining was performed as previously described. (Ennever, et al., Biochem. Biophys. Acta, 826:67 (1985)). Briefly, the grid was subjected to glow discharge prior to staining. A drop of DNA solution was added to the grid, blotted and stained using 0.04% uranyl **acetate**. Column 5, lines 47-55.
- To compact the nucleic acid, the carrier is added to the nucleic acid solution, whereby the carrier disrupts the nucleic acid: solvent interactions allowing the nucleic acid to condense.

Preferably, at least the turbidity of the solution is monitored as the carrier is added, so that a change in state is promptly detected. Once turbidity appears, the state of the DNA may be further analyzed by CD spectroscopy to determine whether the DNA is in the condensed or the aggregated state. (Precipitation should also be detectable with the naked eye.) Preferably, the carrier is added sufficiently slowly to the nucleic acid solution so that precipitation and aggregation are minimized. If precipitation or aggregation occur, a chaotropic salt should be added slowly, and the result again examined by CD spectroscopy. The preferred salt is NaCl. Other chaotropic salts can be used as long as they are tolerated by the animal (or cells) to which they will be administered. Suitable agents include Sodium sulfate (Na_2 SO_4), Lithium sulfate (Li_2 SO_4), Ammonium sulfate ($(NH_4)_2$ SO_4, Potassium sulfate (K_2 SO_4), Magnesium sulfate ($MgSO_4$), Potassium phosphate (KH_2 PO_4), Sodium phosphate (NaH_2 PO_4), Ammonium phosphate (NH_4 H_2 PO_4), Magnesium phosphate ($MgHPO_4$), Magnesium chloride (Mg Cl_2), Lithium chloride (LiCl), Sodium chloride (NaCl), Potassium chloride (KCl), Cesium chloride (CaCl), Ammonium **acetate**, Potassium **acetate**, Sodium **acetate**, Sodium fluoride (NaF), Potassium fluoride (KF), Tetramethyl ammonium chloride (TMA-Cl), Tetrabutylammonium chloride (TBA-Cl), Triethylammoniym chloride (TEA-Cl), and Methyltriethylammonium chloride (MTEA-Cl). Column 21, lines 47-67.

- We have observed variations in the function described by the above equation when we use different DNA plasmids and different DNA preparations during the condensation process. These differences are probably related to the variation in the affinity of poly-L-lysine for DNA of different sources and compositions. For maximum binding affinity we generally use DNA precipitated twice with sodium **acetate** and 2.5 volumes of -40.degree. C.; ethanol (see Methods). We have not found an apparent difference in binding affinity of poly-L-lysine for DNA of different forms (i.e. supercoiled, nicked and linear) and for DNA extracted using anionic exchange chromatography or cesium chloride gradient centrifugation. This may indicate the presence of a contaminant in the DNA preparations from different sources which has poly-L-lysine binding activity, that is eliminated by sequential DNA precipitation. Column 22, lines 35-50.

- Cytochemical identification of macrophages. Cells and tissue sections were stained nonspecific esterase activity, which is relatively specific for mononuclear phagocytes. The cell smears were fixed as described above, and incubated with a filtered solution containing a-naphthyl **acetate** and Fast Blue BB salt for 10 minutes at room temperature. Tissue sections were stained with this solution for 1-3 hours, and counterstained with 0.1% nuclear fast red. Column 32, lines 58-65.

- Preparation of Fab fragments. The isolation and papain digestion of antibodies derived from rabbits immunized with rat secretory component has been described previously. Briefly, polyclonal antibody was isolated from rabbit serum using a Protein A MAPS agarose column

as described by the manufacturer. Isolated immunoglobulin G (2 mg) was treated with 20 .mu.g papain for 12 hours at 37.degree. C. in the presence of 100 mM sodium **acetate** (pH 5.5) 50 mM cysteine, and 1 mM EDTA. The Fab fragment was separated from intact antibody and Fc fragments by Protein A chromatography. An irrelevant Fab (IFab) was generated by papain digestion of IgG from pre-immune rabbit serum. Column 37, lines 60 to column 38, line 4.

c. counterion of polycation molecules

The U.S. Patent and Trademark Office asserts, "Hanson makes DNA/polycation complexes by the same procedure described by instant claim 1." Advisory action at page 2, line 6. Similarly, it asserts, "Because claim 1 is a product-by-process claim and Hanson et al. teaches this process, the examiner asserts that such a rod-shaped complex is formed." *Id.* at lines 14-15. The process of claim 1 recites that the polycation molecules have acetate[11] as a counterion prior to mixing with the nucleic acid molecule.

Hanson teaches at column 25, lines 38-52:

> Polymers of L-lysine-HBr or L-lysine-Cl with an average chain length of 109 (Sigma) were galactosylated essentially as described by Monsigny, et al. (1984) Biol. Cell., 51, 187. Briefly, 2 mg of poly-L-lysine was reacted with 89 g of .alpha.-D-galactopyranosyl phenyl-isothiocyanate (Sigma G-3266) dissolved in N,N-Dimethyl formamide (5 mg/ml). The solution was adjusted to pH 9.0 by the addition of 1/10 volume of 1M sodium carbonate pH 9.0. Since the reaction is 10% efficient, 0.8% of the .epsilon.-NH_3 groups present in the solution are glycosylated. The tube was shielded from light by aluminum foil and mixed for 6 hours at room temperature. The solution was then dialyzed, using Spectra-Por dialysis tubing (Fisher 3500 M.W. cutoff), against 500 ml of 5 mM NaCl buffer for 2 days with frequent changes of buffer (2 changes/day).

And Hanson teaches at column 42, lines 55-67:

[11] the elected species; in this regard claims 26 and 28 and their dependents 30, 31, 34, 35, stand

> Production of the galactosylated poly-L-Lysine. Poly-L-lysine was galactosylated as described (PNAS). Two mg of poly-L-lysine-HBr (Sigma P-7890, average chain length, 100) was reacted with 85 mg of a-D-galactopyranosyl phenyl-isothiocyanate (Sigma G-3266). The solution was adjusted to pH 9 by the addition of 1/10 volume of 1M sodium carbonate pH 9. The tube was shielded from light by aluminum foil and mixed for 16 hours at room temperature, then dialyzed using Spectra-Por dialysis tubing (3500 M.W. cutoff) against 500 ml of 5 mM NaCl for 2 days with frequent changes of buffer (4 changes/day). The reaction is stoichiometric and resulted in the galactosylation of 0.8 to 1% of the NH_3 groups present in the solution.

After dialysis, the resulting polycation polymers had chloride as a counterion. Hanson teaches mixing these polycation/counterion preparations with the nucleic acid molecule. See column 26, lines 12-16 and column 43, 19-24.

2. Martin, *FEBS Letters 480:*106-112 (2000)

Martin is cited as teaching interconversion of toroidal and rod-like DNA complexes. Martin teaches that these are intermediates, and likely not fully condensed products:

> The toroidal structures observed in this system are believed to be fairly loosely wound structures due to the bulky nature of the pegylated-polymer, hence probably represent an early stage of condensate formation. We believe we are observing a stage of condensate formation where ring and rod-like structures exist dynamically, having the ability to reversibly equilibrate between structures.

Page 111, column 1, third full paragraph.

Martin observes his structures using atomic force microscopy (AFM), in which the observed structures are tethered to a solid support:

> AFM has been utilized in a number of applications in the study of DNA, including the visualisation of dynamic processes [16,17] and indeed looking specifically at systems for DNA delivery

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separately from the other claims because they are limited to acetate.

> [8,18,19]....The requirement of immobilization of molecule to substrate does though remain.

Page 106, column 2, paragraph 2.

Martin teaches the use of mica as the solid support to which the complexes attach:

> Condensates were prepared by adding 20 μl of polymer solution (138 μg ml^{-1}) to 20 μl of DNA solution (20 μg ml^{-1}). After 5 min incubation, 20 μl of the resulting solution was deposited onto 1 cm^2 of freshly cleaved mica.

Page 107, column 1, lines 15-18.

E. Differences between claimed subject matter and reference teachings

1. Hanson does not inherently teach the same product as claimed because Hanson does not teach the same process of making.

Hanson does not teach the same process of making because Hanson does not teach use of polycations with an acetate counterion as a reagent for mixing with nucleic acids. Each of the rejected claims requires the use of elected species acetate as a counterion for the polycation prior to mixing.

a. Hanson does not teach acetate as a counterion.

Hanson teaches other counterions, but not acetate. Hanson teaches polycations which initially have bromide or chloride as a counterion.[12] These are dialyzed against sodium chloride, which yields polycations with chloride as a counterion. Hanson teaches mixing the chloride-dialyzed polycations with nucleic acids.

b. <u>Hanson teaches acetate for other purposes.</u>

Each of the mentions of acetate in the Hanson reference is quoted above. None of the mentions are as a counterion for a polycation for mixing with nucleic acid to form a complex. Hanson teaches using uranyl acetate to stain samples for electron microscopy. Column 5, lines 47-55. Hanson teaches using acetate salts as chaotropic salts to add to a precipitated or aggregated complex. Column 21, lines 47-67. Hanson teaches using acetate salts to precipitate naked DNA. Column 22, lines 35-50. Hanson teaches staining tissue sections with a solution comprising acetate. Column 32, lines 58-65. Hanson teaches the use of acetate in a solution for performing an enzymatic reaction on antibodies. Column 37, lines 60 to column 38, line 4.

None of these teaches the use of acetate as a counterion for a polycation that is mixed with nucleic acid molecules to form complexes.

2. Hanson does not explicitly teach the same product as claimed.

Hanson does not describe the same products as claimed. While indeed both Hanson and the subject application describe compositions that are heterogeneous, the subject application's composition has a component that is not present or taught in the Hanson composition. Hanson does not teach a complex which is rod-shaped, 10-20 nm diameter, and contains a condensed nucleic acid.

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[12] Hanson, col. 25, lines 38-52

3. Hanson's toroids do not spontaneously form condensed rods.

The U.S. Patent and Trademark Office points to a post-filing date article by Martin[13] to support an alternative theory of anticipation by inherency. According to this alternative theory, Hanson's toroids would spontaneously convert to rods, because Martin observed conversion of toroids to rods. However, Martin's conditions are quite different from Hanson's. Martin's complexes are adhered to a silica substrate, whereas Hanson's complexes were condensed in bulk solution. These conditions are indeed different, as Martin points out:

> In common with all surface imaging there is the question of how closely the composition and morphology of adsorbed structures represents that in the bulk solution. In this study repeated imaging of the sample was possible over a number of hours. Regardless of the duration of imaging no structures were observed other than the rod-like and toroidal condensates already discussed, though the number of condensates on the substrate increased over time as more complexes diffused to the surface. It would be expected that if other structures, for example spherical aggregates, were present then they would adhere to the substrate in a similar manner to the condensates observed.

Page 111, column 2, lines 18-29.

The intrinsic evidence indeed indicates that Martin's conditions are different than the Hanson conditions, as other complex forms present in Hanson's compositions, such as aggregates, are not formed under Martin's conditions, and the Martin complexes do not progress or resolve beyond the early stage. "We believe we are observing a stage of condensate formation where ring and rod-like structures exist dynamically, having the ability to reversibly equilibrate between structures." Page 111, column 1, third full paragraph. The evidence suggests that Martin's adherence of the complexes to the solid substrate captures early stage condensates in an intermediate stage which does not permit resolution to final products.

[13] *FEBS Letters 480:* 106-112 (2000)

Contrary to the assertion of the U.S. Patent and Trademark Office, Martin does not teach a general phenomenon of dynamic equilibrium between all rod and toroidal complexes under all conditions. Hanson teaches the presence of condensed toroids but not of condensed rods. There is no evidence that the dynamic equilibrium present under Martin's conditions would be in effect under Hanson's conditions. There is no evidence that the Martin conversion would or did occur in the Hanson preparations.

Thus Martin does not demonstrate that the condensed toroids of Hanson inherently convert to form rods of condensed nucleic acid.

F. Conclusion

Hanson does not teach the same process of making as recited. Hanson does not teach the same compositions. Hanson's compositions do not spontaneously form the claimed compositions. Hanson does not anticipate the claimed invention because it neither explicitly nor inherently teaches the recited invention.

II. Hanson, Park, and Schacht do not render claims 3, 10, 19, 31, 51-53, 63-65, 67, 68, 76-78, and 104 obvious under 35 U.S.C. §103(a) Hanson, Park, and Mao do not render claims 58-62, 66, 73-75, 79-82, and 122 obvious under 35 U.S.C. §103(a) Hanson, Park, Schacht, and Kwoh do not render claims 4-5, 13-14, 39-40, 54-55, 69-70, 106-107, 114-115 obvious under 35 U.S.C. §103(a)

A. Legal standard for obviousness

Section 103(a) of 35 U.S.C. states:

A patent may not be obtained though the invention is not

> identically disclosed or described as set forth in section 102 of this title, if the differences between the subject matter sought to be patented and the prior art are such that the subject matter as a whole would have been obvious at the time the invention was made to a person having ordinary skill in the art to which said subject matter pertains.

Obviousness under 35 U.S.C. § 103(a) is a question of law based on several factual inquiries:

> Under § 103, the scope and content of the prior art are to be determined; differences between the prior art and the claims at issue are to be ascertained; and the level of ordinary skill in the pertinent art resolved.

Graham v. John Deere Co., 383 U.S. 1, 17 (1966). In *KSR Int'l v. Teleflex Inc.*, 550 U.S. 398, 407 (2007), the Supreme Court explained, "While the sequence of these questions might be reordered in any particular case, the [*Graham*] factors continue to define the inquiry that controls." The U.S. Patent and Trademark Office bears the initial burden of establishing a *prima facie* case of obviousness based on the results of the factual inquiries under *Graham*. M.P.E.P., 8th ed., § 2142.

It remains black letter law that obviousness requires at least a suggestion of all of the features in a claim. See *CFMT, Inc. v. Yieldup Intern. Corp.*, 349 F.3d 1333, 1342 (Fed. Cir. 2003). As the Supreme Court stated in *KSR*, "there must be some articulated reasoning with some rational underpinning to support the legal conclusion of obviousness." *KSR Int'l v. Teleflex Inc.*, 550 U.S. 398, 418 (2007), citing *In re Kahn*, 441 F.3d 977, 988 (Fed. Cir. 2006). Moreover, the Court affirmed the corollary principle that "when the prior art teaches away from combining certain known elements, discovery of a successful means of combining them is more likely to be nonobvious." *KSR Int'l v. Teleflex Inc.*, at 416. The rejections of all claims as obvious fail because they fail to teach or suggest all features of the claimed invention and because the prior art teaches away from the claimed invention.

B. <u>The cited art</u>

Hanson's teachings have been discussed in detail above. In brief, Hanson does not teach a method of making which utilizes a polycation with an acetate counterion, nor does Hanson teach complexes that are unaggregated, rod-shaped, 10-20 nm in diameter, have condensed nucleic acid, and are colloidally stable in normal saline.

Park is cited for teaching the use of polyethylene glycol (PEG) on polylysine attached through an amino terminal linkage.

Schacht is cited for teaching a disulfide linkage via a cysteine moiety of polylysine to PEG.

Mao is cited for teaching the lyophilization of complexes and administration to cells.

Kwoh is cited for teaching that polylysines of all sizes condense plasmid DNA into toroids and rod-shaped structures as shown by electron microscopy ranging in size from 40 to 80 nm for rods. Further, Kwoh is cited by the U.S. Patent and Trademark Office for teaching that polyethylene glycol (PEG) conjugation to poly-L-lysine (PLL) -DNA makes longer rods and more rods and that size can be measured using electron microscopy.

Kwoh teaches the instability of her complexes in a number of different ways. In Table 1, Kwoh compares the size of her polylysine complexes (PLL10K and PLL26K) in water to the size in 0.15 M NaCl. The complexes in saline have a 5-fold increased particle size. "Immediately after the addition of NaCl (0.15 M final concentration) the effective diameter of PLL polyplexes increased at all charge ratios (Fig. 3) and continued to increase in size with time (data not shown)." Page 177, column 2, lines 10-14. Kwoh teaches that growth in size reflects instability: "PLL and AsOR-PLL polyplexes exhibited colloidal instability which accounts for the drastic increase in size at neutral charge rations of PLL polyplexes." Page 187, column 2, lines 7-9. Similarly, Kwoh teaches that PEG-lysine complexes are not colloidally stable in physiological saline. Complexes made with DNA and PLL10K-PEG5K have a diameter of 80.5 nm in water, which increases to 187 nm in saline (see page 185, column 1, line 12 to column 2, line 3). The increased size indicates instability of the complex in normal saline.

C. Differences between the prior art and the claimed subject matter

Hanson does not teach the recited complexes which have the following properties:

- o unaggregated;
- o rod-shaped;
- o 10-20 nm diameter;
- o condensed nucleic acid component;
- o colloidally stable in normal saline.

Hanson teaches aggregate complexes of increased size (>60 nm). Col. 62, lines 51-53. These are not unaggregated and do not have a diameter of 10-20 nm. Hanson teaches rod-like, relaxed toroids of increased size. Col. 62, lines 53-54. These do not have a condensed nucleic acid component. Hanson teaches properly condensed toroids. Col. 62, lines 54-56. These are not rod-shaped. Hanson also does not teach how to make the recited complexes by using acetate as a counterion to a polycation. The secondary references do not remedy the deficiencies.

Kwoh does not teach remaining elements of the claims. Kwoh does not teach the use of a polycation with an acetate counterion and does not teach the formation of complexes that are colloidally stabile in normal saline. Kwoh teaches complexes that are colloidally unstable. Page 187, column 2, lines 7-9. Kwoh thus teaches away from the claimed invention and provides evidence of non-obviousness.

None of the other cited references teaches or suggests the use of acetate as a counterion for the polycation used to condense nucleic acids. Thus the references in combination fail to teach all elements of the claims.

Using acetate as a counterion, complexes with the recited properties are formed. Compositions which comprise these complexes are colloidally stable as shown in figures 11, 15,

and 18.

D. Level of skill in the art

The level of skill in the art in the area of biochemistry, molecular biology, and gene therapy is high. Nonetheless, these remain areas of substantial unpredictability. There is no evidence of record that indicates that one of skill in the art would have known at the time of the invention that using acetate as the counterion of the polycation used as a reagent to form DNA complexes would have resulted in improved particle properties such as colloidal stability in normal saline, as disclosed in the present invention.

E. Conclusion

Appellants respectfully request that the Board reverse the rejections as the prior art neither teaches nor suggests the subject matter of the claims. Moreover, the cited prior art actually teaches away from the stable complexes of the invention.

Respectfully submitted,

BANNER & WITCOFF, LTD.

Date: May 27, 2009

By: /SARAH A. KAGAN/

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APPENDIX 1. APPEALED CLAIMS

1. A non-naturally occurring composition comprising a plurality of unaggregated nucleic acid complexes, wherein individual complexes of said plurality consist essentially of a single nucleic acid molecule and one or more polycation molecules, wherein said complexes are formed by mixing said nucleic acid molecule and said polycation molecules, wherein prior to mixing said polycation molecules have a counterion selected from the group consisting of acetate, bicarbonate, and chloride, wherein a subset of said complexes are rod-shaped when visualized by transmission electron microscopy, wherein the rod-shaped complexes have a diameter of 10-20 nm when visualized by transmission electron microscopy, wherein the nucleic acid molecules of the rod-shaped complexes are condensed, and wherein said complexes are colloidally stable in normal saline.

2. The composition of claim 1 wherein the polycation molecules are polylysine or a polylysine derivative.

3. The composition of claim 2 wherein the polylysine derivative is polylysine peptide with a cysteine residue.

4. The composition of claim 1, wherein said rod-shaped complexes have a length of 100-300 nm when visualized by transmission electron microscopy.

5. The composition of claim 1, wherein the rod-shaped complexes have a length of 100-200 nm when visualized by transmission electron microscopy.

6-7. (Canceled)

8. A non-naturally occurring composition comprising a plurality of unaggregated nucleic acid complexes, wherein individual complexes of said plurality consist essentially of a single nucleic acid molecule and one or more polycation molecules, wherein said complexes are formed by mixing said nucleic acid molecule and said polycation molecules, wherein prior to mixing said polycation molecules have a counterion selected from the group consisting of acetate, bicarbonate, and chloride, said polycation molecules having a nucleic acid binding moiety through which they are complexed to the nucleic acid, wherein said nucleic acid molecule encodes at least one functional protein, wherein a subset of said complexes are rod-

shaped when visualized by transmission electron microscopy, wherein the rod-shaped complexes have a diameter of 10-20 nm when visualized by transmission electron microscopy, wherein the nucleic acid molecules of the rod-shaped complexes are condensed, and wherein said complexes are colloidally stable in normal saline.

9. The composition of claim 8 wherein the polycation molecules are polylysine or a polylysine derivative.

10. The composition of claim 9 wherein the polylysine derivative is polylysine peptide with a cysteine residue.

11. The non-naturally occurring composition of claim 8 wherein said nucleic acid molecule comprises a promoter which controls transcription of an RNA molecule encoding the functional protein.

12. The non-naturally occurring composition of claim 8 wherein the protein is therapeutic.

13. The non-naturally occurring composition of claim 8 wherein the rod-shaped complexes have a length of 100-300 nm when visualized by transmission electron microscopy.

14. The non-naturally occurring composition of claim 8 wherein the rod-shaped complexes have a length of 100-200 nm when visualized by transmission electron microscopy.

15-16. (Cancelled)

17. A non-naturally occurring composition comprising a plurality of unaggregated nucleic acid complexes, wherein individual complexes of said plurality consist essentially of a single double-stranded cDNA molecule and one or more polycation molecules, wherein said complexes are formed by mixing said nucleic acid molecule and said polycation molecules, wherein prior to mixing said polycation molecules have a counterion selected from the group consisting of acetate, bicarbonate, and chloride, wherein said cDNA molecule encodes at least one functional protein, wherein a subset of said complexes are rod-shaped when visualized by transmission electron microscopy, wherein the nucleic acid molecules of the rod-shaped complexes are condensed, wherein the rod-shaped complexes have a diameter of 10-20 nm when visualized by transmission electron microscopy, and wherein said complexes are colloidally

stable in normal saline.

18. The composition of claim 17 wherein the polycation molecules are polylysine or a polylysine derivative.

19. The composition of claim 18 wherein the polylysine derivative is polylysine peptide with a cysteine residue.

20-25. (Cancelled)

26. A non-naturally occurring composition comprising a plurality of soluble compacted complexes of a nucleic acid molecule and one or more polycation molecules, wherein a subset of said complexes are rod-shaped when visualized by transmission electron microscopy, wherein the rod-shaped complexes have a diameter of 10-20 nm when visualized by transmission electron microscopy, wherein individual complexes of said plurality consist essentially of a single nucleic acid molecule and one or more polycation molecules, wherein the nucleic acid molecules of the rod-shaped complexes are condensed, wherein said complexes are colloidally stable in normal saline, wherein said complexes are made by the process of:

mixing a nucleic acid with a polycation having acetate as a counterion, at a salt concentration sufficient for compaction of the complexes.

27. (Cancelled)

28. A non-naturally occurring composition comprising a plurality of soluble compacted complexes of a nucleic acid molecule and one or more polycation molecules, wherein a subset of the complexes are rod-shaped when visualized by transmission electron microscopy, wherein the nucleic acid molecules of the rod-shaped complexes are condensed, wherein the rod-shaped complexes have a diameter of 10-20 nm when visualized by transmission electron microscopy, wherein individual complexes of said plurality consist essentially of a single nucleic acid molecule and one or more polycation molecules wherein said complexes are colloidally stable in normal saline, wherein the complexes are made by the process of:

mixing a nucleic acid molecule with polycation molecules having acetate as a counterion in a solvent to form a complex, said mixing being performed in the absence of added salt, whereby the nucleic acid forms soluble complexes with the polycation molecules without forming aggregates.

29. (Cancelled)

30. The composition of claim 26 wherein the polycation molecules are polylysine or a polylysine derivative.

31. The composition of claim 30 wherein the polylysine derivative is polylysine peptide with a cysteine residue.

32.-33. (Cancelled)

34. The composition of claim 28 wherein the polycation molecules are polylysine or a polylysine derivative.

35. The composition of claim 34 wherein the polylysine derivative is polylysine peptide with a cysteine residue.

36.-37. (Canceled)

38. The composition of claim 17 wherein the nucleic acid complexes are associated with a lipid.

39. The composition of claim 17 wherein said rod-shaped complexes have a length of 100-300 nm when visualized by transmission electron microscopy.

40. The composition of claim 17 wherein the rod-shaped complexes have a length of 100-200 nm when visualized by transmission electron microscopy.

41.-50. (Cancelled)

51. The composition of claim 1 wherein said polycation molecules are CK15-60P10 and the counterion is acetate, wherein CK15-60P10 is a polyamino acid polymer of one N-terminal cysteine and 15-60 lysine residues, wherein a molecule of polyethylene glycol having an average molecular weight of 10 kdal is attached to the cysteine residue.

52. The composition of claim 51 wherein the polycation molecules comprise 30 residues of lysine.

53. The composition of claim 51 wherein the polycation molecules comprise a targeting moiety.

54. The composition of claim 51, said rod-shaped complexes have a length of 100-300

nm when visualized by transmission electron microscopy.

55. The composition of claim 51, wherein the rod-shaped complexes have a length of 100-200 nm when visualized by transmission electron microscopy.

56-57. (Cancelled)

58. The composition of claim 51 which is lyophilized.

59. The composition of claim 51 which is rehydrated after lyophilization.

60. The composition of claim 51 which does not contain a disaccharide.

61. A method of delivering polynucleotides to cells comprising:

contacting the composition of claim 59 with cells, whereby the nucleic acid is delivered to and taken up by the cells.

62. The method of claim 61 wherein the composition does not contain a disaccharide.

63. The composition of claim 8 wherein the polycation molecules are CK15-60P10, and the counterion is acetate, wherein CK15-60 is a polyamino acid polymer of one N-terminal cysteine and 15-60 lysine residues, wherein a molecule of polyethylene glycol having an average molecular weight of 10 kdal is attached to the cysteine residue.

64. The composition of claim 63 wherein the polycation molecules comprise 30 residues of lysine.

65. The composition of claim 63 wherein the polycation molecules comprise a targeting moiety.

66. The composition of claim 63 which is lyophilized.

67. The non-naturally occurring composition of claim 63 wherein said nucleic acid molecule comprises a promoter which controls transcription of an RNA molecule encoding the functional protein.

68. The non-naturally occurring composition of claim 63 wherein the protein is therapeutic.

69. The non-naturally occurring composition of claim 63 wherein the rod-shaped complexes have a length of 100-300 nm when visualized by transmission electron microscopy.

70. The non-naturally occurring composition of claim 63 wherein the rod-shaped complexes have a length of 100-200 nm when visualized by transmission electron microscopy.

71-72. (Cancelled)

73. The composition of claim 63 which is rehydrated after lyophilization.

74. The composition of claim 63 which does not contain a disaccharide.

75. A method of delivering polynucleotides to cells comprising:

contacting the composition of claim 73 with cells, wherein the polynucleotide encodes a protein, whereby the protein is expressed.

76. The composition of claim 17 wherein said polycation molecules are CK15-60P10, and said counterion is acetate, wherein CK15-60P10 is a polyamino acid polymer of one N-terminal cysteine and 15-60 lysine residues, wherein a molecule of polyethylene glycol having an average molecular weight of 10 kdal is attached to the cysteine residue.

77. The composition of claim 76 wherein the polycation molecules comprise 30 residues of lysine.

78. The composition of claim 76 wherein the polycation molecules comprise a targeting moiety.

79. The composition of claim 76 which is lyophilized.

80. The composition of claim 76 which is rehydrated after lyophilization.

81. The composition of claim 76 which does not contain a disaccharide.

82. A method of delivering polynucleotides to cells comprising:

contacting the composition of claim 80 with cells, wherein the polynucleotide encodes a protein, whereby the protein is expressed.

83.-102. (Cancelled)

103. The composition of claim 8 wherein the nucleic acid complexes are associated with a lipid.

104. The composition of claim 17 wherein the nucleic acid complexes are associated with a lipid.

105. (Cancelled)

106. The composition of claim 26 wherein the complexes have a length of 100-300 nm.

107. The composition of claim 26 wherein the complexes have a length of 100-200 nm.

108-113. (Cancelled)

114. The composition of claim 28 wherein the complexes have a length of 100-300 nm.

115. The composition of claim 28 wherein the complexes have a length of 100-200 nm.

116.-121. (Cancelled)

122. The method of claim 75 wherein the composition does not contain a disaccharide.

123. (Cancelled)

APPENDIX 2. EVIDENCE RELIED UPON

none

APPENDIX 3. RELATED PROCEEDINGS

There are no related court or Board proceedings. A sibling divisional application is pending, S. N. 10/307555.